The opinion in support of the decision being entered today was not written for publication and is not binding precedent of the Board.

UNITED STATES PATENT AND TRADEMARK OFFICE

BEFORE THE BOARD OF PATENT APPEALS AND INTERFERENCES

Ex parte JACQUES FASTREZ, DANIEL LEGENDRE, and PATRICE SOUMILLION

Appeal No. 2006-0300
Application No. 08/978,607

HEARD March 7, 2006

MAILED

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U.S. PATENT AND TRADEMARK OFFICE
BOARD OF PATENT APPEALS
AND INTERFERENCES

Before ELLIS, ADAMS, and GRIMES, Administrative Patent Judges.

GRIMES, Administrative Patent Judge.

DECISION ON APPEAL

This appeal involves claims to an analytical method using modified enzymes. The examiner has rejected the claims as nonenabled and lacking adequate written description. We have jurisdiction under 35 U.S.C. § 134. We affirm the nonenablement rejection and do not reach the written description rejection.

Background

The specification discloses "chimeric target molecule[s] having an activity which can be regulated or modulated by a binding molecule." Page 1. "The term 'chimeric' target molecule, e.g., a 'chimeric enzyme,' means the resultant product after [a] binding site moiety has been inserted into the target molecule or after a portion of the target

molecule has been replaced by the binding site moiety." Page 3. "[A] mimotope is the preferred BSM [binding site moiety]." Page 2.

A "mimotope" is defined with reference to a particular binding molecule: "A mimotope is a determinant which is recognized by the same binding molecule as a particular 'epitope' but which has a different composition from the 'epitope.' For example, a binding molecule can be an antibody which recognizes (i.e. binds to) an epitope comprising a linear sequence of amino acids. A 'mimotope' of this epitope comprises a different linear sequence of amino acids but which is still recognized by the same antibody." Page 4.

"The binding site moiety can be engineered into any desired position in the target molecule. . . . Preferably, as discussed above, the resultant chimeric TM [target molecule] retains at least some of its activity after engineering of the BSM. In addition, attachment of a BM [binding molecule] to the BSM results in regulating the aforementioned activity of the chimeric target molecule. . . . Thus, a preferred site of engineering, e.g., insertion, is a position where the activity of the TM is not eliminated but which, when replaced or modified by the addition of amino acid residues, can act as a regulatory switch for TM activity." Pages 10-11.

"Upon attachment of the BM to the BSM, an activity associated with the TM is altered in a detectable way, e.g., increasing or reducing the activity of the TM." Page 2. "The effect of the binding molecule can be to reduce or even eliminate the activity. . . . The binding molecule can also affect activity in other ways, e.g., increase it, change its specificity, activate it, etc." Page 17.

"A chimeric target molecule can be used to detect the presence or amount of an analyte in test sample. . . . The chimeric enzyme is contacted with a (1) test sample containing an analyte, and (2) a substrate upon which the chimeric TM enzyme catalytically acts, to form a reaction mixture. . . . Binding or attachment of the antibody or BM to the chimeric enzyme can modulate catalysis on the substrate by the chimeric enzyme." Pages 20-21. Or "the activity of a reaction mixture, comprising a chimeric enzyme and a binding molecule (BM) which modulates the activity of the chimeric enzyme, can be further affected by an analyte (a ligand of the binding molecule). The analyte can act as a direct competitor of the interaction of the chimeric enzyme with BM: addition of the analyte competes or displaces the binding molecule from TM, reversing its modulatory effect on the detectable activity." Page 21.

Discussion

1. Claim construction

Claims 13-23 and 25-29 are on appeal. Claims 24, 30-35, 37 and 38 are also pending: the examiner has indicated that claims 30-35, 37 and 38 are allowable and has objected to claim 24 as dependent on a rejected base claim.

Claims 13 and 20 are the only independent claims on appeal and read as follows:

> 13. A method for determining the presence or amount of an analyte in a test sample, comprising:
>
> forming a mixture of (1) a chimeric enzyme comprising a starting enzyme and a mimotope, said mimotope including at least one amino acid, said chimeric enzyme having a sequence of said mimotope inserted in said enzyme or replacing at least one amino acid of said enzyme with the proviso that the activity of the chimeric enzyme is modulated upon binding of a binding molecule to the mimotope, (2) a test sample, (3) a binding molecule which binds to a mimotope of the chimeric enzyme and

modulates the activity of the enzyme, and (4) a substrate upon which the chimeric enzyme catalytically acts; and

detecting the amount of catalysis of the substrate and thereby determining the presence or absence of said analyte of interest.

20. A method for determining the presence or amount of an analyte in a test sample, comprising:

forming a mixture of (1) a chimeric enzyme comprising a starting enzyme and a mimotope, said mimotope including at least one amino acid, wherein said chimeric enzyme having a sequence of said mimotope inserted in said enzyme or replacing at least one amino acid or said enzyme with the proviso that the activity of the chimeric enzyme is modulated upon binding of a binding molecule to the mimotope, (2) test sample, and (3) a substrate upon which the chimeric enzyme catalytically acts; and

detecting the amount of catalysis of the substrate and thereby determining the presence or absence of said analyte of interest.

Thus, claim 13 is directed to an assay for detecting an analyte that interferes with binding of a binding molecule (BM) to a mimotope that has been inserted into a chimeric enzyme: if the analyte is present in the sample, it will interfere with binding of the BM to the mimotope and thereby decrease the effect of the BM on the chimeric enzyme.

Claim 20 is directed to an assay for detecting an analyte (BM) that itself binds to the mimotope: if the analyte is present in the sample, it will bind to the mimotope and modulate the activity of the chimeric enzyme.

Both claims also implicitly require that the chimeric enzyme used in the method include a mimotope that binds the BM of interest. That is, when the claims are read in light of the specification, they are directed to a method of using a chimeric enzyme that has been screened for binding to a specific BM in order to assay for the presence of an analyte in a sample.

2. Enablement

The examiner rejected claims 13-23 and 25-29 under 35 U.S.C. § 112, first paragraph, on the basis that the specification does not enable those skilled in the art to practice the full scope of the claimed method without undue experimentation. The examiner acknowledged that the specification enables those skilled in the art to make and use chimeric enzymes based on β-lactamase but concluded that the guidance in the specification relating to β-lactamase could not be extrapolated to other enzymes without undue experimentation. See the Examiner's Answer, pages 6-7.

The examiner reasoned that the effect of inserting a mimotope into a starting enzyme (other than β-lactamase) will produce unpredictable results "because every other enzyme is distinct in its sequence, regions of active site or susceptibility to modifications." Id., page 7. The examiner noted that the specification provides no working examples of chimeric enzymes "other than β-lactamase modified at the specific positions" (id.), and lacks guidance regarding "where the sequence inserts of the mimotope (BSM) can be made, identification of the active catalytic and binding sites and the effect(s) of such modifications on the functionality of the different enzyme constructs." Id., page 8. Without such guidance, the examiner concluded, undue experimentation would be required to practice the full scope of the claims. Id.

"When rejecting a claim under the enablement requirement of section 112, the PTO bears an initial burden of setting forth a reasonable explanation as to why it believes that the scope of protection provided by that claim is not adequately enabled by the description of the invention provided in the specification of the application." In re Wright, 999 F.2d 1557, 1561-62, 27 USPQ2d 1510, 1513 (Fed. Cir. 1993). "[T]o be

enabling, the specification of a patent must teach those skilled in the art how to make and use the full scope of the claimed invention without 'undue experimentation.'" Id. at 1561, 27 USPQ2d at 1513.

"Whether undue experimentation is needed is not a single, simple factual determination, but rather is a conclusion reached by weighing many factual considerations." In re Wands, 858 F.2d 731, 737, 8 USPQ2d 1400, 1404 (Fed. Cir. 1988). Those factors include "(1) the quantity of experimentation necessary, (2) the amount of direction or guidance presented, (3) the presence or absence of working examples, (4) the nature of the invention, (5) the state of the prior art, (6) the relative skill of those in the art, (7) the predictability or unpredictability of the art, and (8) the breadth of the claims." Id.

A. Breadth of the claims

Claims 13 and 20 are directed to analytic methods using "a chimeric enzyme comprising a starting enzyme and a mimotope, said mimotope including at least one amino acid, said chimeric enzyme having a sequence of said mimotope inserted in said enzyme or replacing at least one amino acid of said enzyme with the proviso that the activity of the chimeric enzyme is modulated upon binding of a binding molecule to the mimotope." Thus, the claims require using an enzyme that meets the following criteria:

(1) It is modified compared to a naturally occurring (starting) enzyme;

(2) The modification consists of insertion of an amino acid sequence or replacement of part of the naturally occurring amino acid sequence with another amino acid sequence (i.e., a mimotope); and

(3) The mimotope is located in the chimeric enzyme such that the enzyme's activity is altered (modulated) when the binding molecule binds the mimotope.

The specification discloses that chimeric enzymes can be based on any of several enzymes that have been previously displayed on phage (paragraph bridging pages 5 and 6) and other classes of enzymes (page 6, first full paragraph). More generally, however, the specification states that "the chimeric TM can be any molecule having a desired activity, e.g., enzymatic." Page 23. Thus, the claims read on a method of using a chimeric enzyme based on any starting enzyme.

The specification states that "[b]inding of the BM to the chimeric molecule, preferably at the BSM, can affect activity in various ways. The binding molecule can inactivate the chimeric TM. By the term 'inactivate,' it is meant that the activity of the chimeric TM is reduced or weakened." Page 7. "The activation of a chimeric molecule can also be regulated by a BM. The simplest example of activation is the proteolytic cleavage of a peptide bond in a zymogen to transform it into an enzyme. . . . An antibody, or other BM, binding to an epitope or mimotope engineered into the region of the cleaved peptide bond can inhibit the activation." Page 9.

"Preferably, . . . the resultant chimeric TM retains at least some of its activity after engineering of the BSM. In addition, attachment of a BM to the BSM results in regulating the aforementioned activity of the chimeric target molecule. . . . Thus, a preferred site of engineering, e.g., insertion, is a position where the activity of the TM is not eliminated but which, when regulated or modified by the addition of amino acid residues, can act as a regulatory switch for TM activity." Pages 10-11.

Although the specification describes as "preferred" a chimeric enzyme that retains at least some activity after insertion of a mimotope, we consider that to be a requirement of the chimeric enzyme used in the method of claims 13 and 20: the

specification provides no guidance on how to make or use a chimeric enzyme that completely lacks activity following insertion of a mimotope.

B. Guidance and working examples

The specification states that "[t]he site where a BSM is engineered, e.g., inserted into and/or replaced, in the TM can be selected by various ways as the skilled worker would know. For example, if the three-dimensional (3D) structure of the TM is known, a site can be selected by specifically identifying a desired location on the molecule to engineer. . . . If the 3D-structure is not known, the site of engineering can be selected on the basis of other information, e.g., when the structure of the protein is not known, sites susceptible to limited proteolysis or sites strongly predicted to be loops by secondary structure prediction or by analysis of hydrophobic patterns are suitable for engineering. . . . Alternatively, a BSM can be engineered at random positions within the TM." Page 11.

The specification provides working examples that describe insertion of DNA fragments encoding random sequences of amino acids, into a gene encoding a fusion of β-lactamase and the phage fd coat protein pIII. See page 26. The specification states that the mimotope insertion sites were chosen based on their positions relatively close to the active site and their poor sequence conservation among β-lactamases. See pages 28 and 36. These are apparently relevant factors for enzymes that have known 3D structures (Figure 1C shows the 3D structure of β-lactamase) and related family members with which their sequences can be compared. The exemplified libraries were shown to contain chimeric enzymes in which activity was modulated when the mimotope was bound by an antibody. See pages 55-56.

The specification provides the following guidance with regard to where mimotopes should be inserted in enzymes generally:

> The engineered site is preferably not at the active site, more preferably it is at a location remote from it, e.g., about 1, 5, 15, 20, or 25 Å from it. The activity of the chimeric molecule must be regulatable by binding to the inserted or replaced sequence, irrespective of whether the modification is close or remote from the active site.

Pages 11-12.

C. Nature of the invention, predictability, and the state of the prior art

The claimed method requires a chimeric enzyme that has a mimotope inserted in such a way that the chimeric enzyme retains some of the activity of the starting enzyme and also shows a detectable change in enzyme activity when the mimotope is bound by its cognate binding molecule. The evidence of record shows that obtaining these two properties simultaneously in a chimeric enzyme involves a high degree of unpredictability.

For example, Brennan[1] and Benito[2] disclose chimeric enzymes made by inserting a known epitope into a starting enzyme. Both references started with enzymes having known 3D structures and inserted epitopes into the enzyme at positions expected to have specific properties. See Brennan, page 512, right-hand column ("[T]he crystal structure of AP provides . . . information that was the basis for choosing sites in alkaline phosphatase that are close to the active site and that might accommodate insertions of peptides without destroying the general structure and enzymatic activity of the protein."); Benito, page 21251, right-hand column ("We have inserted the main antigenic region . . .

[1] Brennan et al., "Modulation of enzymatic activity by antibody binding to an alkaline phosphatase-epitope hybrid protein," Protein Engineering, Vol. 7, pp. 509-514 (1994).
[2] Benito et al., "β-galactosidase enzymatic activity as a molecular probe to detect specific antibodies," Journal of Biological Chemistry, Vol. 271, pp. 21251-21256 (1996).

in different zones of β-gal [β-galactosidase] which are important for the stabilization of the active site.").

Brennan discloses that inserting a 13- or 15-amino acid peptide into alkaline phosphatase at either of two positions produced chimeric enzymes having enzymatic activity. See Table 1. Brennan also discloses that an antibody specific to the inserted epitope bound each of the chimeric enzymes, referred to as API1, API6, and API7. Paragraph bridging pages 511 and 512. However, Brennan discloses that antibody binding affected activity in only one of the three chimeric enzymes produced. See page 511, right-hand column: "The activities of wild type AP, API6 and API7 are not significantly altered by the α-gp120 mAb. In contrast, API1 enzymatic activity is inhibited by α-gp120 mAb." Brennan also teaches that insertions at a third location "resulted in proteins that were degraded in vivo." Page 512, right-hand column. Finally, Brennan presents two possible mechanisms for antibody inhibition of the API1 chimera and states that either or both mechanisms could be involved, along with a variety of other factors. See page 513.

Benito discloses that inserting a 27- and/or 57-amino acid peptide into the enzyme β-galactosidase at either or both of two positions produced chimeric enzymes having enzymatic activity. See Table I. All of the chimeric enzymes were bound by an antibody to the inserted peptide. See page 21253, left-hand column ("Although the constructions gave different reactivities, all of them recognized the mAbs."). Benito discloses that antibody binding enhanced enzymatic activity of some, but not all, of the chimeric enzymes. See page 21253, left-hand column: "[P]rotein M278VP1 is clearly activated by interaction with both mAbs. The activity increased with increasing

concentrations of mAb up to about 200%. Proteins AB1 and M275SVP1 presented an enhancement of about 150%. . . . On the other hand, the presence of mAbs had no [ ] significant effect on the activity of M275VP1 and AB1275VP1."

Benito teaches that even though the crystal structure of β-galactosidase was known, and the sites of insertion were chosen as "zones . . . important for the stabilization of the active site" (abstract), the effect of the insertions on enzyme activity was not predictable before they were made. See page 21254, right-hand column: "It seemed likely that insertions performed at these regions might not only combine the enzymatic activity with a good antigenicity of the peptide, but also <u>inhibit or activate</u> β-gal upon antibody binding by sterically blocking or altering the structure of the active site" (emphasis added). Benito presents two hypothetical mechanisms to explain the observed results. See page 21255, left-hand column.

Thus, the prior art Brennan and Benito references show that the effect of inserting an antigenic peptide into an enzyme was unpredictable, even when the enzyme was well-characterized and the sites of insertion were chosen based on the same criteria disclosed in the specification. In addition, the prior art shows that antibody binding to a chimeric enzyme produced unpredictable effects, sometimes increasing or decreasing activity and sometimes having no effect at all.

<u>D. Undue experimentation</u>

In light of the factual considerations reviewed above, we conclude that the evidence of record supports the examiner's conclusion that undue experimentation would have been required to practice the full scope of the claimed method. The effect of inserting an antigenic peptide into an enzyme was unpredictable, as was the effect on

enzyme activity of binding an antibody to a chimeric enzyme. The specification offers little guidance to the skilled worker, applicable to enzymes generally, regarding where to place an antigenic peptide in a starting enzyme in order to predictably obtain the properties required for the chimeric enzymes used in the claimed method.

Practicing the claimed method throughout its scope, therefore, would involve extensive trial-and-error experimentation by the skilled worker. The experimentation would include making different chimeric enzymes and testing them to determine which retained enzymatic activity, testing to determine which of the active chimeras were bound by a particular binding molecule, and testing to determine which of the active, antibody-binding chimeras had activities that were modulated by antibody binding.

It is true that some experimentation, even a considerable amount, may not be "undue" if it is merely routine, or if the specification provides a reasonable amount of guidance as to the direction in which the experimentation should proceed. See In re Wands, 858 F.2d 731, 737, 8 USPQ2d 1400, 1404 (Fed. Cir. 1988). In this case, however, the claims encompass methods using chimeric enzymes based on any starting enzyme; the process of making the chimeric enzymes required to practice the claimed method involves many sources of unpredictability; and the specification offers little guidance as to the direction the experimentation should proceed. We therefore conclude that the evidence of record supports the examiner's conclusion that practicing the full scope of the claims would have required undue experimentation.

E. Appellants' arguments

Appellants argue that the claims are directed to a method, not a genus of chimeric enzymes, and that those skilled in the art can use the disclosed β-lactamase

chimeric enzymes to screen for any desired mimotope. Therefore, Appellants conclude, the chimeric β-lactamase enzymes disclosed in the specification are sufficient to enable practice of the full scope of the claimed method. See the Appeal Brief, pages 11-14; Reply Brief, pages 3-8.

We do not agree with Appellants' reasoning. The claims on appeal are not limited to a method of using chimeric enzymes based on β-lactamase, they encompass methods of using chimeric enzymes based on <u>any</u> starting enzyme. Enablement requires a disclosure that allows a skilled worker to practice the full scope of the claimed invention without undue experimentation. <u>See</u> <u>In re Wright</u>, 999 F.2d 1557, 1561, 27 USPQ2d 1510, 1513 (Fed. Cir. 1993). ("[T]o be enabling, the specification of a patent must teach those skilled in the art how to make and use <u>the full scope of the claimed invention</u> without 'undue experimentation.'") (emphasis added); <u>In re Vaeck</u>, 947 F.2d 488, 496, 20 USPQ2d 1438, 1445 (Fed. Cir. 1991) ("[T]here must be sufficient disclosure, either through illustrative examples or terminology, to teach those of ordinary skill in the art how to make and how to use the invention <u>as broadly as it is claimed</u>.") (emphasis added).

The claims encompass methods of screening using chimeric enzymes based on starting enzymes other than β-lactamase. Therefore, the claims are not necessarily enabled simply because methods of using chimeric β-lactamase enzymes are enabled. For the reasons discussed in detail above, we conclude that the evidence supports the examiner's conclusion that practicing the full scope of the claimed methods would have required undue experimentation.

Appellants also argue that, "should the skilled artisan want to use an enzyme other than β-lactamase in the claimed methods, the specification provides the needed guidance." Appeal Brief, page 17. Appellants point to the specification's teaching of particular enzymes that could be used and the "identifi[cation of] three factors to consider when modifying a starting enzyme" (id.). Appellants argue that "[t]ools for evaluating all three factors were well known to those skilled in the art at the time the application was filed." Id., page 18 (citing a journal article, "the BlastP program," and an on-line protein data bank). Appellants argue that the amino acid sequence and location of "catalytic and binding sites" of enzymes other than β-lactamase are "well-known, well characterized and readily available to one of ordinary skill in the art, based on the description of starting enzymes in the specification." Reply Brief, page 19.

Appellants' argument does not persuade us that the claimed methods could be practiced throughout their scope without undue experimentation. First, we have considered the limited guidance provided by the specification with regard to where a mimotope should be inserted into a starting enzyme to obtain a chimeric enzyme having the desired properties. The three factors cited in the specification do not substantially reduce the amount of experimentation required to practice the full scope of the claims.

First, the specification provides no working examples or other evidence to show that chimeric enzymes will have the desired properties if they have mimotopes inserted anywhere other than in nonconserved regions near the active site, as in the working examples. As discussed above, the Benito and Brennan references show that the effect of inserting an epitope (or mimotope) in an enzyme is unpredictable. There is no evidence in the record to indicate that inserting a mimotope at "an exposed site on the

surface of the target molecule" or "sites susceptible to limited proteolysis or sites strongly predicted to be loops" (specification, page 11) would be likely to result in a chimeric enzyme having the properties required by the claims.

Second, Appellants have cited no evidence in the record to support their argument that tools were readily available for identifying the active site and nonconserved regions of enzymes, as well as numerous 3D protein structures. The assertions on page 18 of the Appeal Brief are attorney argument, not evidence, and Appellants have not pointed out where the record shows that the cited reference and Web pages were considered by the examiner. The same applies to Appellants' assertions that enzyme amino acid sequences and active site locations were "readily available to one of ordinary skill in the art." Reply Brief, page 19.

A preponderance of the evidence of record supports the examiner's position that undue experimentation would have been required to practice the full scope of the claimed method. Appellants have not adequately rebutted the examiner's prima facie case. We therefore affirm the rejection of claims 13 and 20 for failure to meet the enablement requirement of 35 U.S.C. § 112, first paragraph. Claims 14-19, 21-23, and 25-29 fall with claims 13 and 20 because Appellants did not argue any claims separately. See 37 CFR § 41.37(c)(1)(vii).

3. Written description

The examiner also rejected claims 13-23 and 25-29 under 35 U.S.C. § 112, first paragraph, on the basis that the specification does not adequately describe the claimed methods, with emphasis on the chimeric enzymes used in the methods. Since we have

already concluded that these claims are nonenabled, we need not reach the issue of whether they are adequately described.

Other Issue

During prosecution, the examiner rejected claims 13 and 20 as anticipated by Benito and Brennan. See the Office action mailed April 1, 2003. These rejections were withdrawn in the Examiner's Answer in view of "Applicants' arguments and amendments to the claims." See page 3. Thus, the examiner has apparently concluded that these references do not identically disclose the claimed method.

The examiner has not, however, addressed on the record whether Benito or Brennan would have made obvious any embodiment within the scope of claims 13 and 20. Cf. In re Slayter, 276 F.2d 408, 411, 125 USPQ 345, 347 (CCPA 1960) ("It is well settled that a generic claim cannot be allowed to an applicant if the prior art discloses a species falling within the claimed genus."). We note in this regard that the "mimotope" recited in the claims need only vary in a single amino acid from the corresponding, naturally occurring epitope. See the specification, page 4 ("The 'mimotope' differs by at least one amino acid from the 'epitope.'").

In In re Mayne, 104 F.3d 1339, 41 USPQ2d 1451 (Fed. Cir. 1997), the court addressed the obviousness of a compound that differed from the prior art only by the substitution of a single amino acid: the claimed compound had the sequence Phe-Pro-Leu where the prior art taught Phe-Pro-Ile. See id. at 1343, 41 USPQ2d at 1454. The court concluded that the claimed compound would have been obvious because of the structural similarity between the known and claimed compounds: "Structural relationships often provide the requisite motivation to modify known compounds to

obtain new compounds. In fact, Leu is an isomer of Ile – an identical chemical formula with differences only in the chemical bonding of the atoms. . . . The structure of Leu and Ile alone suggest their functional equivalency." Id., 41 USPQ2d at 1454-55 (citation omitted).

Since a mimotope can apparently differ from its corresponding epitope by substitution of Leu for Ile, or some other conservative amino acid substitution, if this application or a related application comes before the examiner again, he should consider whether the claimed subject matter would have been obvious in view of the prior art, under the reasoning relied on by the court in In re Mayne.

Summary

We affirm the rejection of claims 13-23 and 25-29 based on the enablement requirement of 35 U.S.C. § 112, first paragraph.

AFFIRMED

J Ellis
Joan Ellis
Administrative Patent Judge

Donald E. Adams
Donald E. Adams
Administrative Patent Judge

Eric Grimes
Administrative Patent Judge

BOARD OF PATENT APPEALS AND INTERFERENCES

EG/dym

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